

# The Toxicologic Effects of the Carbamate Insecticide Aldicarb in Mammals: A Review

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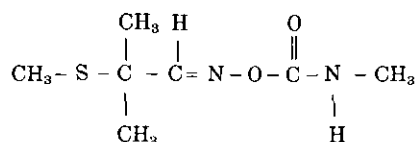
Aldicarb, 2-methyl-2-(methylthio)propionaldehyde-*O*-methylcarbamoyloxime, is an oxime carbamate insecticide manufactured by the Union Carbide Corporation and sold under the trade name Temik. It is a soil-applied systemic pesticide used against certain insects, mites, and nematodes, and is applied below the soil surface for absorption by plant roots. It is generally applied to the soil in the form of 5, 10, or 15% granules, and soil moisture is essential for the release of the toxicant. Uptake by plants is rapid. Aldicarb is currently registered for use on cotton, sugar beets, sugar cane (Louisiana only), potatoes, sweet potatoes, peanuts, oranges, pecans (Southeast only), dry beans, soybeans, and ornamental plants. Home and garden use is not permitted.

Discovery of aldicarb and its oxidative sulfoxide and sulfone metabolites in well or ground water in Florida, Wisconsin, and New York, and accidental poisonings from ingesting contaminated watermelons and cucumbers in the South and West have spurred interest and concern about this pesticide.

The primary mechanism of toxic action of aldicarb is cholinesterase inhibition. However, unlike the relatively irreversible anticholinesterase activity of the organophosphate pesticides, the carbamylation process which produces the anti-AChE action is quickly reversible. Aldicarb is readily absorbed through both the gut and the skin, but is rapidly metabolized and excreted in the urine almost completely within 24 hr. Although it is acutely toxic to humans and laboratory animals, aldicarb is not known to be carcinogenic, teratogenic, conclusively mutagenic, or to produce other long-term adverse health effects. In cases of accidental poisoning, the cholinergic symptoms have generally subsided within 6 hr, with no side effects or complications.

## Introduction

Aldicarb, 2-methyl-2-(methylthio)propionaldehyde-*O*-(methylcarbamoyl)-oxime, is an oxime carbamate insecticide produced by the Union Carbide Corporation under the code number "UC 21149," trademark "Temik," and has the structural formula:



Aldicarb is relatively soluble in water and most organic solvents. It is a stable compound, except in strong alkaline solutions, and is nonflammable and noncorrosive to metal.

Aldicarb is a systemic pesticide used against a variety of insects and nematodes. The insecticide is applied below the soil surface for absorption by plant root systems, and moisture is required to release the toxicant from granules. Aldicarb is currently registered for use on a variety of crops, including cotton, sugar beets, sugar

cane, certain citrus fruits, potatoes, sweet potatoes, peanuts, and ornamental plants; it may generally provide plant protection for a period of up to 12 weeks after application, depending on soil and climatic conditions. The half-life of aldicarb varies from approximately 1 to 2 weeks under laboratory conditions, but may be much shorter in the field or longer if it leaches to groundwater or if cool, nonalkaline conditions prevail.

## Absorption, Metabolism, and Excretion

While it is not the intent of this article to provide an in-depth presentation of the metabolism of aldicarb and its pathways in man and animals, the authors feel that a brief discussion of this area would facilitate the understanding of the toxicological properties and health impacts of this insecticide.

A number of studies have demonstrated that aldicarb, as well as its sulfoxide and sulfone metabolites, is absorbed readily and almost completely through the gut in a variety of species (1-5). Dermal absorption has been reported in rabbits (6,7) and rats (8) and might be expected to present a problem to unprotected humans in manufacturing and field application settings.

The rapidity of gastrointestinal absorption is illustrated in a study by Cambon et al. (5), who administered

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by gastric intubation single doses of 0.001, 0.01, and 0.10 mg aldicarb (analytical grade 99%) per kg body-weight to pregnant Sprague-Dawley rats. Signs of poisoning occurred approximately 5 min after the administration of the larger doses, indicating the speed of uptake from the gut.

The extent of absorption, metabolism, and excretion of aldicarb has also been documented in number of other studies. Andrawes et al. (2) administered oral doses of radiolabeled Temik and Temik sulfoxide to female rats and observed 80 to 90% excretion of the isotope in the urine and 2 to 5% in the feces within the first 24 hr following administration. Only trace amounts of Temik were found in the excreta, attesting to the high degree of absorption. Another study provided further evidence of the rapidity and completeness of aldicarb absorption (1). Following oral administration of Temik labeled in three separate positions, the researchers reported that Temik was rapidly absorbed from the gastrointestinal tract. By the end of the first 24 hr after administration, approximately 90% of two of the labels had been excreted in the urine or as CO<sub>2</sub>. When rats were fed milk containing radiolabeled Temik metabolites daily for 9 days, approximately 90% of each dose was reported to have been excreted in the urine within a 24-hr period (3). Similarly, Dorrough et al. (4) fed Holstein cows various concentrations of Temik in the diet and found that 92, 3, and 1% of the dosages were eliminated in the

urine, feces, and milk, respectively, indicating almost complete absorption of the administered dose through the gut.

Studies examined in the preparation of this review indicate that aldicarb is widely distributed throughout the contaminated organism. However, since it is rapidly metabolized, invasion of individual tissues is generally temporary, given noncontinuous exposure.

Andrawes et al. (2) administered oral doses of radiolabeled S<sup>35</sup> Temik to female rats at a level of 0.4 mg/kg. At designated times (5–11 days following treatment), two rats were killed and tissue samples were removed and frozen for future analysis. Tissues analyzed included blood, heart, kidneys, brain, lungs, spleen, liver, leg muscle, fat, bone, stomach, and small and large intestine. Analysis of the tissue samples indicated a general distribution of radioactive residues, and there was no indication that any particular tissue or group of tissues was selective in sequestering aldicarb or its metabolites.

The metabolism of aldicarb involves both hydrolysis of the carbamate ester and oxidation of the sulfur to the sulfoxide and sulfone derivatives (2). While the hydrolysis results in compounds with little or no insecticidal activity or toxicity to other organisms, the sulfoxide and sulfone metabolites are active cholinesterase inhibitors (2,9,10). Figure 1 shows the metabolic pathways of aldicarb in the rat (11). Aldicarb metabolites in

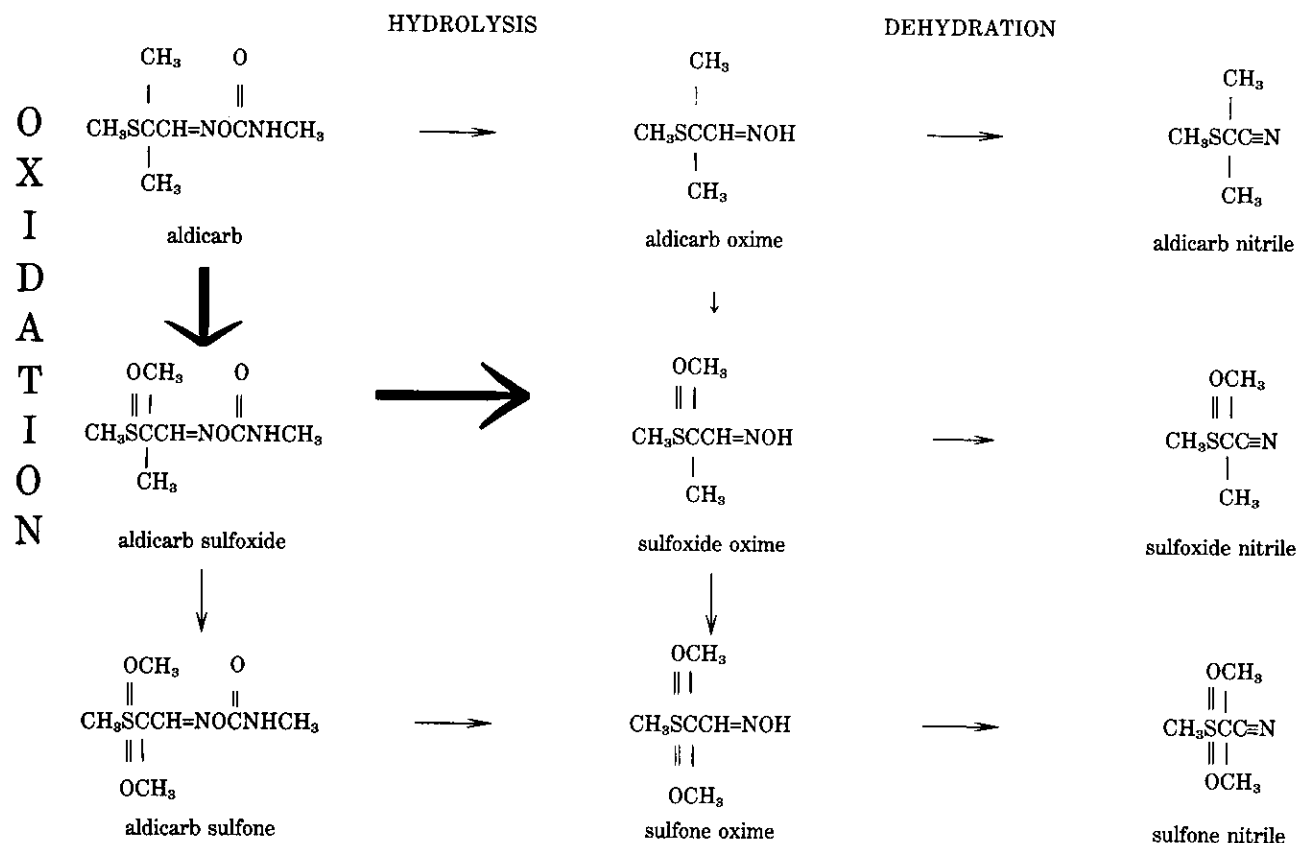


FIGURE 1. Metabolic pathways of aldicarb metabolism in rats. Source: Modified from Wilkinson et al. (11).

animals include aldicarb sulfoxide, aldicarb sulfone, oxime sulfoxide, oxime sulfone, nitrile sulfoxide, nitrile sulfone, and at least five other metabolites (1,4).

Further investigation of the metabolism of aldicarb was reported in a study of the metabolic fate of the insecticide Temik labeled at three different positions (either S-methyl-C<sup>14</sup>, t-butyl-C<sup>14</sup>, or N-methyl-C<sup>14</sup>) (1). In this experiment, male Carworth Farms-Elias stock rats were administered 50 µg oral doses (equivalent to 0.33 mg/kg body weight) of radiolabeled Temik dissolved in polyethylene glycol 400 using the weighed syringe technique. Each labeled compound was administered to eight animals. In addition, groups of four rats per radiolabel were similarly administered 10 mg (66 mg/kg bodyweight) of the S-methyl-C<sup>14</sup> and t-butyl-C<sup>14</sup> oxime analogues. Results of this study indicated that Temik is metabolized in the rat and excreted in the first-day urine as Temik sulfoxide (~40%), oxime sulfoxide (~30%), and 5 to 9 other polar compounds (~30%), believed to be acids. Essentially all of the test compounds administered were metabolized by the rat. Only trace quantities of Temik (~1%) and of oxime and oxime sulfone (~1-2%) could be found in the urine. Evidence of Temik sulfone could not be found by either gas, silica gel, or thin layer chromatography.

The metabolism of aldicarb has also been studied experimentally in cows (3). The results of this study indicated that the metabolic pathway of the insecticide Temik in dairy animals is similar to that reported for other animal species. Additional detailed reports of the specific metabolic pathways for aldicarb are available in the published literature (2,12-14).

As previously discussed, the elimination of aldicarb and its metabolites has been studied in a variety of species including rats (1-3,15) and cows (3,4). In all studies reviewed, the primary means of aldicarb excretion was found to be via the urine. Respiratory CO<sub>2</sub> and feces are other minor avenues through which this insecticide and its metabolites are eliminated from exposed animals. Enterohepatic recirculation of aldicarb and/or its metabolites has been demonstrated by Marshall and Dorrough (15), who suggested that in intact rats (study performed with cannulated bile ducts), the majority of the biliary metabolites are reabsorbed from the intestine and ultimately excreted in the urine.

## Summary

Aldicarb is readily absorbed through the gut and skin. It is rapidly metabolized and excreted within 24 hr of exposure, with urine accounting for almost all of the excreted toxic and relatively nontoxic (oxime and nitrile) metabolites.

## Health Effects Studies Using Animals

### Acute Toxicity

The National Academy of Science reports that the acute toxicity of aldicarb is probably the highest of any

widely used insecticide (10). Table 1 provides LD<sub>50</sub> values for aldicarb in a variety of species.

The acute toxicity of this insecticide has been studied in a variety of laboratory mammals, including rats (8,16-21), mice (22), and rabbits (18).

Gaines (8) administered technical grade aldicarb as single doses in peanut oil via stomach tube or dermally as a solution in xylene to adult Sherman rats of both sexes. Female rats were found to be slightly more sensitive to Temik than were males. Oral LD<sub>50</sub> values were determined to be 0.8 and 0.65 mg/kg body weight for males and females, respectively. Reported dermal LD<sub>50</sub> values were 3.0 mg/kg body weight for males and 2.5 mg/kg for females. The lowest doses reported to have killed rats were 0.5 mg/kg body weight for oral administration and 2.0 mg/kg body weight for the dermal route for animals of both sexes. Other oral LD<sub>50</sub> studies produced similar, but slightly higher, values in rats (see Table 1). LD<sub>50</sub> values for aldicarb sulfoxide and aldicarb sulfone were determined to be 0.88 mg/kg body weight and 25.0 mg/kg, respectively, in another study in rats (19).

When aldicarb was administered orally using a vehicle other than peanut or corn oil, the LD<sub>50</sub> values were somewhat higher. Weil (20) found the acute oral LD<sub>50</sub> for Temik 10G to be 7.07 mg/kg body weight in rats

Table 1. Acute toxicity of aldicarb.

LD <sub>50</sub>	Route of administration	Organism/strain	Vehicle	Reference
0.93 mg/kg	Oral	Rats		(7,16)
0.8 mg/kg (males)	Oral	Sherman rats	Peanut oil	(8)
0.65 mg/kg (females)	Oral	Sherman rats	Peanut oil	(8)
0.9 mg/kg (males)	Oral	Rats (unspecified type)	Corn oil	(17)
1 mg/kg (females)	Oral	Rats (unspecified type)	Corn oil	(18)
3 mg/kg (males)	Dermal	Sherman rats	Xylene	(8)
2.5 mg/kg (females)	Dermal	Sherman rats	Xylene	(8)
0.3-0.5 mg/kg	Oral	Swiss white mice		(22)
5.0 mg/kg	Dermal	Rabbits	Propylene glycol (5% solution)	(16)
5.0 mg/kg	Dermal	Rabbits		(18)
32 mg/kg <sup>a</sup>	Dermal	Rabbits	Water (50% solution)	(23)
12.5 mg/kg <sup>b</sup>	Dermal	Rabbits	Dimethyl phthalate	(18,23)
3.5 mg/kg <sup>b</sup>	Dermal	Rabbits	Toluene (5% solution)	(18,23)
141 mg/kg <sup>a</sup>	Dermal	Rabbits	None <sup>c</sup>	(24,25)
0.44 mg/kg	IP	Rats (unspecified type)		(21)
1.60 mg/kg <sup>d</sup>	Oral	Deer mice		(26)

<sup>a</sup> 24-hr exposure.

<sup>b</sup> 4-hr exposure.

<sup>c</sup> Dry Temik 10G applied to skin under adhesive tape.

<sup>d</sup> Reported as approximate lethal dose (ALD), not an LD<sub>50</sub> value.

when the insecticide was administered as dry granules. Carpenter and Smith (17) placed 1 g of Temik 10G in 200 mL of water for 8 hr and then administered the formulation to rats. The resulting LD<sub>50</sub> was determined to be 6.2 mg/kg body weight.

In addition to the study by Gaines (8) mentioned previously, the dermal toxicity to rats has been investigated in at least two other studies. Carpenter and Smyth (17) conducted 4-hr skin exposure tests with rats by using Temik 10G applied as both dry formulation and with saline added. Following the 4-hr exposure period, in which the test materials were taped to the skin, the dry formulation was found to be not as toxic as was the same mixture when wetted with saline. The respective LD<sub>50</sub> values were 400 mg/kg body weight for the moistened mixture and 3200 mg/kg for the dry application. Weil (20), in a similar experiment, applied dry granules of Temik 10G or Temik 15G to the bellies of rats for 4 hr and found the LD<sub>50</sub> values to be 4.58 and 6.32 g/kg body weight for the 10% (10G) and 15% (15G) formulations, respectively. The significance of the specific vehicle in dermal applications of aldicarb is evident in Table 1.

In inhalation studies using rats, mice, and guinea pigs, a concentration of 200 mg aldicarb dust/m<sup>3</sup> was found to be highly toxic to all three species in a 5-min exposure (17). At this level, rats and mice were observed to be more susceptible to the dust than were guinea pigs. The survival at a given concentration was found to vary with time. While rats appeared to be capable of surviving an aldicarb dust concentration of 6.7 mg/m<sup>3</sup> for 15 min, five of six were killed by the same concentration in 30 min. By contrast, rats were able to survive 8 hr in atmospheres of saturated aldicarb vapors. It appears that aldicarb is somewhat less toxic as an aerosol than a dust, as attested by the observation that two of six rats were able to survive an 8 hr exposure to a 7.6 mg aldicarb/m<sup>3</sup> aerosol. When the concentration of the aerosol was raised to 15 mg/m<sup>3</sup>, five of six animals exposed died within 4 hr.

A much larger number of short-term studies have been conducted using higher doses and a broader variety of test species. Weil and Carpenter reported a 93-day study (27) in which aldicarb was administered in the diet of Catworth Farms-Elias male and female rats at dosages of 0, 0.02, 0.1, and 0.5 mg/kg/day. A significant increase in mortality (cause not specified) was noted at the high dose; mortality was further reported to be numerically, but not significantly, increased at the 0.1 mg/kg/day level. On examination of pathology, the organ weights and plasma, erythrocyte, or brain cholinesterase levels in survivors at all dosage levels were found not to differ from controls. When aldicarb was fed at a level of 3.2 mg/kg/day, body weights of males and females were depressed, while liver and kidney weights were not. Plasma cholinesterase activity was depressed in both males and females, but a reduction in erythrocyte cholinesterase activity was observed only in males.

In a shorter duration study with rats (28), the effects of aldicarb, aldicarb sulfoxide, and aldicarb sulfone were examined on groups of 10 (5 males and 5 females each) rats. Commencing when the rats were 45 days of age, the groups were administered the test compound in the feed for 7 days at dosages including 0.4, 1.0, 2.5, or 5.0 mg/kg/day. Parameters investigated included effects on body weight, liver and kidney weights, and brain, plasma and erythrocyte cholinesterase activity. Rats were said to tolerate > 0.4 mg aldicarb sulfoxide/kg/day without effects on body weight, liver and kidney weight, or depression of brain, plasma, or erythrocyte cholinesterase. Aldicarb sulfone was found to depress brain, plasma, and erythrocyte cholinesterase at 5.0 mg/kg body weight. A statistically significant decrease in brain cholinesterase was observed in female rats administered the 2.5 and 1.0 mg/kg body weight dosages. Only limited information was available for this study (40).

DePass et al. reported the administration of a 1:1 mixture of aldicarb sulfoxide and aldicarb sulfone in drinking water to young Wistar rats (29). Groups consisting of 10 animals of each sex were provided water containing nominal concentrations of 19.2, 4.8, 1.2, 0.3, 0.075, or 0.0 ppm of the treatment mixture. Individual dosage levels, based on the nominal concentrations used, and initial weights of the animals, ranged from 0 to 1.67 mg/kg/day for males and from 0 to 1.94 mg/kg/day for females. Blood samples were taken after 8, 15, and 29 days of treatment and were analyzed for plasma and erythrocyte cholinesterase levels. Brain cholinesterase determinations were made at sacrifice (29 days after initiation of treatment).

Plasma cholinesterase was reduced 65 to 77% (depending on the sex of the animal and the day of blood sampling) in all animals at the 19.2 ppm dosage. The 4.8 ppm treatment level of the sulfoxide-sulfone mixture was reported to cause a statistically significant (28% mean) reduction in plasma cholinesterase only in male rats and only on one sampling date, after 8 days of treatment.

RBC cholinesterase activity was likewise reduced at all sampling periods in all 19.2 ppm-treated rats, with reported decreases ranging from 48 to 63%, again depending on the sex of the animal examined and the date of the sample. A reduction of 24% erythrocyte cholinesterase was further observed in males at the 4.8 ppm dose level following 29 days of exposure. No inhibition was observed at any of the lower concentrations.

The only reported statistically significant reduction in brain cholinesterase was a 10% decrease in female rats following 29 days of treatment at the highest dosage level. Males experienced a similar decrease in brain cholinesterase activity, but, because of the greater variability of the data, the reduction was not found to be statistically significant. No mortality or symptoms of acetylcholinesterase inhibition were observed.

Body weight gain and water consumption were also reported to be reduced significantly in both sexes at 7,

14, 21, and 29 days into treatment, as was food consumption during the same time in males; however, the authors indicated that the body weight effect may have been the result of lower palatability of the test solution at the highest concentration, rather than a toxic effect.

In consideration of true toxic effects in this study, it should be noted that the authors reported that the mean concentration of sulfoxide plus sulfone was only approximately 86% of the nominal values when the solutions were first given to the rats, a phenomenon attributed to degradation occurring prior to administration to the animals. Additionally, after 1 week in water bottles at room temperature, the mean concentrations were further reduced to only 73% of the nominal values (which might mean that the observed effects were actually seen at lower than the nominal dosages). The authors concluded that, while substantial effects resulted from exposure of the rats to the 19.2 ppm nominal dosage, the treatment effects of reduced plasma and erythrocyte acetylcholinesterase activity in males at the 4.8 ppm dosage (0.47 mg/kg body weight) during only 1 of the 3 sampling periods were of questionable biological significance, and therefore termed the concentration of 4.8 ppm in the drinking water a "no-ill-effect level" for aldicarb residue exposure in the rat.

It was further noted that the 24 to 28% reduction in acetylcholinesterase activity observed at this dosage generally does not cause symptoms of intoxication in humans (30). If the observed reduction in plasma and erythrocyte cholinesterase activity in the 4.8 ppm males were to be considered an adverse or ill effect, however, the no-observed-adverse-effect level (NOAEL) would be considerably lower (1.2 ppm, or 0.12 mg/kg body weight). It should be noted that the use of the 1:1 mixture of aldicarb sulfoxide and aldicarb sulfone in this study was based on a 1983 Cornell University report (11), which noted that residues found in drinking water are generally present in a 1:1 mixture of sulfoxide to sulfone. Given the known higher toxicity of the sulfoxide moiety, a lower NOAEL might be expected from similar administration of the sulfoxide alone.

Dorough et al. administered Temik in the diet of lactating Holstein cows in a two-part experiment (4). In a pilot study to determine the effect of Temik on the general health of a cow, a single 645 kg cow was given Temik and Temik sulfone at a rate equivalent to 1.2 ppm Temik (0.042 body weight) in the diet for 10 days. Daily blood samples were drawn 6 hr after the morning milking and immediately assayed for RBC and plasma cholinesterase activity. The animal was milked at 12-hr intervals. The results of this pilot study indicated that feeding a dairy cow 1.2 ppm Temik equivalents would not cause any visible ill effects to the animal. The results of these assays indicated that blood cholinesterase levels were not reduced by the treatment, and quantitative milk production remained constant during the treatment.

In the second part of the same study, Dorrough and his co-workers fed three cows separate dosages of a

Temik-Temik sulfone combination for 14 days. The treatment dosages were administered twice daily with a balling gun at 12-hr intervals in the form of a gelatin capsule containing crushed grain. Individual doses of Temik equivalents were 0.12 ppm (0.006 mg/kg body weight) for one cow, 0.6 ppm (0.027 mg/kg body weight) for the second cow, and 1.2 ppm (0.052 mg/kg body weight) for the third. As in the pilot study, blood was taken from each animal on a daily basis throughout the treatment period. The blood was drawn 6 hr after the morning milking and immediately assayed for plasma and RBC cholinesterase activity. All animals were milked regularly at 12-hr intervals. In this experiment, no apparent harmful effects were noted in any of the test animals. Regardless of the level of administered dose, blood cholinesterase levels were the same both before and during treatment. Milk production, feed consumption, and quantity of excretory products were reported to have remained stable throughout the experiment. The authors concluded that, with only minor exception, the continuous exposure of dairy animals to Temik in the diet does not significantly alter the compound's fate, compared to single exposure.

Carpenter and Smyth reported the dosing of cats with 0.5, 1.0, or 1.5 mg aldicarb/kg/day for multiple doses, with a 7 to 8 day interval between doses (17). These authors reported no evidence of tolerance through continued exposure, as death following administration of the third dose was as prompt as the death observed after administration of the first dose.

The potential severity of acute toxic poisoning by aldicarb is evidenced by a reported incident (31) in which six cows became ill and two of the six died following the accidental spilling of Temik in a pasture. Chemical analysis for aldicarb in the rumen of one of the dead animals showed that a lethal dose of the pesticide was present. Milk produced by the surviving animals on the day of the poisoning and the next 6 successive days had to be destroyed to prevent possible human health risks.

The toxic effects of short-term dermal exposure to aldicarb have also been investigated (24,25,32). Carpenter and Smyth investigated the toxic effects of aldicarb administered dermally for a period of 15 days (24). In this study, Temik 10G (10.5% active granular formulation) was administered to male albino rabbits by application of wetted gauze to abraded skin areas for 6 hr/day through a consecutive 15-day period. Dosage levels of 0.05, 0.10, and 0.20 g/kg were tested. Criteria used to assess the toxicity of Temik were total weight gain, food consumption, liver and kidney weights, plasma and erythrocyte cholinesterase activity, and histopathological examination of liver, lung, kidney, heart, muscle, thyroid, and skin tissue. A course of hematology was also run, as were kidney function (blood urea nitrogen) and liver function (serum glutamic oxalacetic and pyruvic transaminase) tests. The only parameter significantly affected was body weight gain, as depressed gains were observed in the groups administered 0.1 and 0.2 g Temik/kg. Plasma cholinesterase activity

was also reported to be depressed at these treatment levels.

### Chronic and Subchronic Effects

Although a number of feeding studies of 3 months or less duration have been conducted (4,17,27,28), only five long-term (chronic) studies could be located in the available literature (33-37). Table 2 summarizes the data from available long-term and significant short-term studies.

Weil and Carpenter (33) studied the effects of aldicarb administered in the diet of rats of an unspecified variety over a 2-year period. Four groups of rats, each group consisting of 20 males and 20 females, were maintained on diets containing aldicarb at levels of 0.005, 0.025, 0.05, or 0.1 mg/kg for 2 years. A control group of the same size was fed a similar diet without the aldicarb. Concurrently, 32 rats per group (16 of each sex) were fed each diet for observation of shorter term effects. In this concurrent experiment, four animals of each sex were sacrificed following the sixth month of the experiment, and the remaining 12 rats of each sex were killed at the end of 1 year. Based upon measurements of food consumption, mortality and lifespan, incidence of infection, liver and kidney weights as percentage of body weight, body weight gain, maximum body weight, hematocrit, incidence of neoplasms, incidence of pathological lesions, and brain, plasma, and erythrocyte cholinesterase levels, the aldicarb-treated animals were found not to differ significantly from controls for any of these

parameters. The results of the 2-year feeding portion of this study are reported later in this paper.

Another long-term feeding study in rats has been reported by Weil and Carpenter (34). In this experiment, groups of 20 Greenacres Laboratory Controlled Flora rats of each sex were fed levels of 0.3 mg aldicarb/kg/day, 0.3 or 0.6 mg aldicarb sulfoxide/kg/day, 0.6 or 2.4 mg aldicarb sulfone/kg/day, or 0.6 or 1.2 mg of a 1:1 mixture of aldicarb sulfoxide and aldicarb sulfone/kg/day for up to 2 years. No adverse effects were observed at the 0.3 mg aldicarb/kg/day treatment level. However, the high dose of the aldicarb sulfoxide, aldicarb sulfone mixture resulted in increased mortality within the first 30 days, as well as a decrease in plasma cholinesterase activity and decreased weight gain in males. Some increase in mortality was also reported for females receiving the 0.6 mg/kg/day aldicarb sulfoxide treatment. No-observed-adverse-effect levels determined in this study were 0.3 mg/kg/day for aldicarb, 0.3 mg/kg/day for aldicarb sulfoxide, 2.4 mg/kg/day for aldicarb sulfone, and 0.6 mg/kg/day for the 1:1 mixture of aldicarb sulfoxide and aldicarb sulfone.

Weil and Carpenter reported the results of a 2-year study in which beagle dogs were administered aldicarb (99.9% pure) in the diet (35). Four groups of dogs, three males and three females each, were fed dosages of aldicarb equivalent to 0, 0.1, 0.05, or 0.025 mg/kg/day. Based upon observations of body weight changes, appetite, mortality, histopathology, hematology, biochemistry, and terminal liver and kidney weights, there were no statistically significant deleterious effects found at any of the administered dosages. The no-effect level of

Table 2. Long-term studies of aldicarb and its oxidative metabolites.

Dose, mg/kg/day	Duration	Route of administration	Organism/strain	Effects	Reference
0.1 <sup>a</sup>	2 years	Oral	Rats (unspecified strain)	None	(33)
0.1	13 weeks	Oral	CFE rats	No adverse effects	(27)
0.5	13 weeks	Oral	CFE rats	Significant increase in mortality	(27)
0.3 <sup>a</sup>	2 years	Oral	Greenacres Laboratory controlled rats	No adverse effects	(34)
0.3	2 years	Oral (aldicarb sulfoxide)	Greenacres Laboratory controlled rats	No adverse effects (some deaths in females at 0.6 mg/kg/day dosage)	(34)
2.4 <sup>a</sup>	2 years	Oral (aldicarb sulfone)	Greenacres Laboratory controlled rats	No adverse effects	(34)
0.6	2 years	Oral (1:1 aldicarb sulfoxide: sulfone mixture)	Greenacres Laboratory controlled rats	No adverse effects (increased mortality within 30 days at 1.2 mg/kg/day)	(34)
0.1 <sup>a</sup>	2 years	Oral	Beagle dogs	None	(35)
0.125	6 months	Oral (aldicarb sulfoxide)	Rats (unspecified strain)	None	(36)
0.12	29 days	Oral (drinking water-1:1 sulfoxide/sulfone ratio)	Wistar rats	None	(29)

<sup>a</sup> Highest dose administered.

0.1 mg/kg/day for dogs is identical to the 0.1 mg/kg/day level determined for rats in the Weil and Carpenter experiment (33) described previously, as well as in 90-day dog studies (38). It should be noted, however, that neither of these Weil and Carpenter studies tested dosages above the 0.1 mg/kg/day level.

In studies using dosages more suitable for the analysis of cholinesterase inhibition (36,37), Weil and Carpenter administered aldicarb sulfoxide or aldicarb sulfone in the diet to rats at dosage levels of 0, 0.125, 0.25, 0.5, or 1.0 mg sulfoxide/kg body weight or 0, 0.2, 0.6, 1.8, 5.4, or 16.2 mg sulfone/kg body weight. Groups of 15 animals of each sex were fed these aldicarb metabolites for a period of 6 months. Animals were sacrificed following 3 or 6 months of treatment for tissue examination and measurement of brain, plasma, and RBC cholinesterase activity. The results of both the 3- and 6-month studies clearly indicated a substantial reduction (relative to controls) of cholinesterase levels at the three highest dosage levels for each compound when measured immediately upon cessation of feeding. The relative order of cholinesterase decrease was plasma > RBC > brain. No mortality or gross or microscopic tissue effects were noted, despite the reported substantial decrease in cholinesterase levels; however, some growth retardation was reported at the highest treatment dosages.

Using similar dosing regimens of aldicarb sulfoxide and sulfone for a 3-month treatment period, Weil and Carpenter (36,37) investigated cholinesterase activity depression in animals either sacrificed immediately upon cessation of feeding or placed on a control diet during a 1-day recovery period. The results of this study indicated a rapid recovery from the cholinesterase inhibition for all but the highest dosage levels for both test compounds. These studies have been used by the World Health Organization Committee on Pesticide Residues to arrive at a 0.125 mg/kg body weight NOEL for aldicarb sulfoxide in the rat (39).

## Carcinogenicity

Aldicarb has been found to be noncarcinogenic to mice in a skin painting study (41) and noncarcinogenic to mice and rats in feeding studies (33,42) at the dosages tested. A summary of these negative studies is presented below.

Weil and Carpenter conducted a skin painting study using male C3H/H3J mice (41). Female mice were not employed for this test due to the incidence of spontaneous mammary tumors, particularly in breeding females. Mice were administered aldicarb in the form of applications of a 0.125% concentration to hair-free skin on the backs of the animals twice per week for up to 28 months. When compared with controls (positive control group painted with cholanthrene), aldicarb was determined to be noncarcinogenic under the conditions of the experiment.

In a 2-year feeding study (33), equivalent doses of 0.005, 0.025, 0.05, or 0.1 mg aldicarb/kg/day were ad-

ministered in the diet to an unspecified strain of rats. The incidences of tumors were determined not to be significantly greater than those of control groups. No adverse effects were also reported in Greenacres Laboratory Controlled Flora rats fed 0.3 mg/kg/day for 2 years (34), although it is not clear whether this particular experiment was designed specifically as an oncogenicity study.

The National Cancer Institute (NCI) conducted a bioassay of aldicarb for possible carcinogenic effects in rats and mice (42). Test groups of 50 male and 50 female F344 rats, and the same numbers of male and female B6C3F1 mice were administered doses of either 2 or 6 ppm in a diet containing 2% corn oil for 103 weeks. The animals were then observed for an additional 0 to 2 weeks and killed. Matched controls were 25 untreated rats and 25 untreated mice of each sex. No tumors that could be clearly determined to be the result of the aldicarb administration occurred in either the rats or mice.

In the NCI bioassay, dose-related adenomas or carcinomas of the pituitary were observed in female rats ( $p = 0.048$ ); however, the incidences of these tumors were not significantly higher than those observed in the corresponding control group. Similarly, fibrosarcomas or sarcomas of subcutaneous tissue were observed in male mice at incidences that appeared to be dose related ( $p = 0.043$ ), but these tumor incidences were not significantly higher than in the corresponding control group. It should be noted that the incidence of pituitary tumors in historical-control female F344 rats at the laboratory conducting the bioassay was also high. Consequently, the occurrence of the pituitary tumors in female rats and subcutaneous tissue tumors of male mice could not be clearly related to the aldicarb administration. Since no tumors occurred at statistically significant levels in either the rats or mice tested, it was concluded that under the conditions of the bioassay, technical grade (99 + %) aldicarb was not carcinogenic for F344 rats or B6C3F1 mice of either sex.

Quarles et al. tested a number of pesticides, including aldicarb, for transforming and tumorigenic activity (43). The purity of aldicarb was indicated to be greater than 95%. In the host-mediated assay portion of this study, pregnant hamsters were given IP injections of 0.001 or 0.005 mg aldicarb/kg bodyweight administered in a non-transforming solvent. Injections were given on day 10 of gestation, and fetal cell cultures were prepared on day 13. In evaluation of this test, colonies were considered to have been transformed if the cells had clearly lost orientation and were criss-crossed or multilayered. When cultures were plated for plating efficiency and determination of transformation, cells were also tested for growth in 0.3% agar.

To test for tumorigenicity, approximately  $10^6$  transformed or untransformed cells were injected SC into young adult nude (athymic) mice. These 4- to 6-week-old mice were then maintained in groups of 3 to 6 per standard polycarbonate cage and observed for 6 to 12 months for tumor formation.



To test for spontaneous transformation, fetal cell cultures were handled at intervals throughout the study and examined according to standard test procedures. Of nearly 38,000 colonies examined, none revealed evidence of morphological transformants, and no growth of cells in 0.3% agar was observed. Therefore, the spontaneous transformation rate was determined to be insignificant in this assay. The solvents used in this experiment were similarly tested, and of approximately 22,000 colonies examined, no induction of morphological transformation or growth of cells suspended in 0.3% agar was observed following treatment of pregnant hamsters with the test solvents.

For the transplacental host-mediated assay, dosages chosen (0.001 and 0.005 mg/kg body weight) were the greatest nonlethal doses tested that did not exceed 0.3 mg/kg bodyweight. Aldicarb showed no transforming activity at the dosages tested. It produced no growth at 0.005%, but was not tested at 0.001 mg/kg. When clones isolated from subcultures 5 to 7 were tested for tumorigenicity in nude mice, none of the four mice inoculated developed tumors. The researchers concluded that aldicarb was inactive and induced neither morphological transformants nor cells that grew in agar. Similar tests with nitrosoaldicarb produced contrasting results, but only aldicarb will be discussed in this paper.

## Mutagenicity

Although conclusive evidence concerning the mutagenic potential of aldicarb is not yet available, two recent studies suggest that this insecticide might have mutagenic potential. In the first study, Cid and Matos investigated the effects of aldicarb on human lymphocytes *in vitro* in the presence of an exogenous metabolic activation system by means of an analysis of sister-chromatid exchange (SCE) and mitotic delay (44). In this study, cyclophosphamide (CP) was used as a positive control to compare the chromosomal effects of aldicarb with a known genotoxic agent. Data obtained from this study revealed that aldicarb, as well as CP, induced a significant increase of SCE values in the absence of S9 mix, leading the authors to suggest that metabolic activation of aldicarb seems to occur in human blood cultures. Treatment with aldicarb in the presence of the S9 mix was reported to induce SCE values slightly higher than those obtained with aldicarb alone. This, the authors state, could be construed to indicate a weak activation of aldicarb by the exogenous metabolic system. Furthermore, S9 mix was reported to apparently potentiate the cytotoxic effects of aldicarb, as evidenced by the observation of only a few metaphases in blood cultures treated with the highest dose of aldicarb alone; no metaphases were observed in cultures treated with the same concentration of aldicarb plus S9.

In the second study (45), Sharaf et al. investigated chromosomal aberrations in male albino rats. The animals received IP injections of 0.00121, 0.00666, or 0.0121 mg/kg body weight of aldicarb in a 1:1 acetone and water vehicle. Controls received only the ace-

tone:water solution. Subacute (1 injection per day for 5 days) and acute (sacrifice 6, 24, or 48 hr following a single injection) phases of the experiment were conducted. Bone marrow cells of rat femurs were analyzed, and increases in both structural and numerical aberrations were observed in all animals treated with aldicarb. Significant structural chromosomal aberrations were observed in the form of chromatid breaks and gaps, centric fusions, centromeric attenuation, and end to end association, suggestive of chromosomal damage occurring during the  $G_1$  stage of the cell cycle. The numerical aberrations were primarily due to polyploidy and endomitosis, the latter indicating a specific effect of the insecticide on the mitotic spindle, and leading to the observed concomitant increase in the mitotic index at all stages. The highest increase in numerical aberrations was observed following subacute treatment with the maximum tolerated dose. The authors suggested that aldicarb may have a cumulative effect and may be a clastogenic agent in rats.

In contrast, U.S. EPA (46) cited an unpublished report (46) indicating that 0.7 mg aldicarb/kg body weight of rats caused no adverse effects in the dominant lethal mutagenicity test. Erecgovich and Rashid evaluated 70 pesticides and their related degradation products for mutagenic effects in five strains of *Salmonella typhimurium*, employing the procedures of the Ames test, and found aldicarb to be weakly mutagenic only without activation by liver microsomal enzymes (47). Belvins et al. (48), Seiler (49), and others have conducted studies using nitrosoaldicarb, but discussion of this chemical is beyond the scope of this review.

## Teratogenicity and Reproductive Effects

Of the limited studies available on the subject, none indicated any teratogenic effects associated with exposure to aldicarb.

The potential of aldicarb as a teratogen was investigated in rats by Weil and Carpenter (50). Pregnant rats were administered dosages of 0, 0.04, 0.20, and 1.0 mg aldicarb/kg body weight in the diet. The rats were further divided into three groups: Group 1 rats were fed aldicarb in the diet throughout pregnancy or until the pups were weaned; Group 2 rats were administered aldicarb from the day the vaginal plug first appeared through the seventh day; and Group 3 received aldicarb from days 5 through 15 of gestation. No congenital malformations were reported for any of the treated groups, and body weights of both the mothers and pups were normal. Although the 1.0 mg/kg dose level was at or near the reported single dose  $LD_{50}$  value for rats (8,16-18), no significant effects were observed concerning fertility, gestation, viability of offspring, or lactation.

In a study conducted by the International Research and Development Corporation (51), groups of 16 pregnant Dutch Belted rabbits each were administered single daily doses of aldicarb to determine its teratogenic potential. Treatment levels of 0, 0.1, 0.25, or 0.50 mg/kg/day were applied via gavage to the respective groups



on days 7 through 27 of gestation. Fetuses were removed surgically by Cesarean section on day 28 of gestation and examined for teratologic effects. One spontaneous abortion each was reported in the 0.25 and 0.5 mg/kg/day groups. Although viable fetuses and total implantation values were less for all treatment groups than for controls, the values fell within historical control ranges. No meaningful differences in fetal malformations or developmental variations between the control and treated groups were reported.

The effects of aldicarb on reproductive performance in rats were investigated in a three-generation study (50), in which the insecticide was incorporated into the diet of the parent generation animals at levels of 0.05 or 0.1 mg/kg/day for 84 days prior to mating. Similar dosages were fed to subsequent generations. Progeny (112 days old) were mated and their offspring used as parents for the  $F_3$  generation pups. Comparison of indices for fertility, gestation, viability, lactation, mean weight of pups, and micropathology of  $F_3$  generation weanlings and 90-day-old  $F_3$  adults revealed no statistically significant differences relative to controls, nor was food consumption affected by either dosage level.

More recently, Cambon et al. (5,52) examined fetal and maternal brain, blood, and liver AChE activity in rats following administration of aldicarb. In the earlier study (5), Cambon and his co-workers administered analytical grade (99%) aldicarb to nulliparous pregnant female Sprague-Dawley rats (3 groups of 8 each) at dosage levels of 0.001, 0.01 or 0.1 mg/kg body weight on day 18 of gestation. Treatments consisted of single doses administered by gastric intubation, and the animals were killed by decapitation 1, 5, or 24 hr following treatment. Blood, brain, and liver samples of both dams and fetuses were analyzed for AChE activity.

Table 3 reflects the results of this study. Signs of poisoning were manifested within 5 min of the 0.1 mg/kg administration. At this dosage level, all of the tissue samples taken 1 and 5 hr following treatment displayed a significant decrease in AChE activity. A decrease in acetylcholinesterase activity was still evident in both maternal and fetal blood 24 hr after administration of

the higher dosage levels. It should be noted, as the authors pointed out, that AChE activity was inhibited in the fetus just 1 hr following a dosage 1000 times lower than the  $LD_{50}$  for aldicarb in the adult. This decrease was noted in the blood, brain, and liver of the fetus, although in adults, only the liver manifested a decrease in AChE activity following administration of this 0.001 mg/kg dose. Acetylcholinesterase activity in the fetus was significantly lowered at all dosages of aldicarb. Further, in a number of instances, the inhibition of AChE activity was greater in fetal than maternal tissue, leading the authors to suggest possible accumulation in fetal tissues.

In a subsequent study (52), Cambon et al. administered by gastric intubation a single dose of 0.1 mg aldicarb/kg body weight to six nulliparous pregnant Sprague-Dawley rats on day 18 of gestation. The rats were killed by decapitation 1 hr after treatment. The acetylcholinesterase enzymes from the brains of mothers and fetuses were separated by electrophoresis on a polyacrylamide gel and stained. The stain intensity of the various electrophoretic fractions was then measured using a densitometer. A significant inhibition of both maternal and fetal cerebral AChE was observed, as was the case in their 1979 experiment. The existence of three distinct isoenzymes was noted. The aldicarb treatment resulted in a significant lowering of the least mobile of the three (isoenzyme 1) in the dams. The levels of isoenzymes 1 and 2 were decreased in the fetuses. The authors suggested that the greater inhibition of fetal (when compared with maternal) brain AChE activity in their 1979 experiment might be due to binding of the inhibitors in the fetus for two of the isoenzymes, while the same dose only decreases the first isoenzyme in the mother. The authors concluded that this study tended to support the hypothesis that the sensitivity difference observed in maternal and fetal AChE to insecticides is in the specific affinities of carbamate derivatives such as aldicarb for each fetal and maternal isoenzyme.

## Health Effects in Humans

Symptoms reported for accidental or occupational poisoning (53-55) and controlled aldicarb human exposure

Table 3. Activity of acetylcholinesterase in maternal and fetal tissues from rats given aldicarb by gastric intubation.\*

Dose, mg/kg	Time after dosing, hr	AChE units/g protein (mean $\pm$ SD)		AChE units/g of organ (mean $\pm$ SD)			
		Maternal blood	Fetal blood	Maternal brain	Maternal liver	Fetal brain	Fetal liver
0	— <sup>b</sup>	22.9 $\pm$ 1.47	24.0 $\pm$ 2.42	9.41 $\pm$ 1.45	5.89 $\pm$ 0.86	1.45 $\pm$ 0.06	1.75 $\pm$ 0.07
	—	23.7 $\pm$ 1.06	17.7 $\pm$ 1.48	9.30 $\pm$ 1.70	3.70 $\pm$ 1.18	1.39 $\pm$ 0.10	1.35 $\pm$ 0.11
0.001	1	22.9 $\pm$ 1.46	21.1 $\pm$ 1.53*	8.93 $\pm$ 2.64	4.88 $\pm$ 0.82*	1.18 $\pm$ 0.15†	1.47 $\pm$ 0.12†
0.01	1	22.0 $\pm$ 2.48	17.9 $\pm$ 5.50‡	8.02 $\pm$ 2.00	4.22 $\pm$ 0.49†	1.00 $\pm$ 0.10†	1.33 $\pm$ 0.31†
	5	18.7 $\pm$ 2.40‡	13.2 $\pm$ 1.90‡	9.50 $\pm$ 1.30	4.20 $\pm$ 0.80‡	1.30 $\pm$ 0.17*	1.50 $\pm$ 0.10†
	24	20.4 $\pm$ 3.63*	13.2 $\pm$ 2.22*	8.16 $\pm$ 2.13	3.13 $\pm$ 0.72	1.19 $\pm$ 0.24*	1.03 $\pm$ 0.15†
0.10	1	16.0 $\pm$ 3.73†	12.7 $\pm$ 1.93†	6.99 $\pm$ 1.84*	2.81 $\pm$ 0.61†	0.83 $\pm$ 0.09†	1.95 $\pm$ 0.19†
	5	15.3 $\pm$ 2.20†	14.5 $\pm$ 2.08†	7.48 $\pm$ 0.82‡	3.55 $\pm$ 0.77†	1.07 $\pm$ 0.06†	1.55 $\pm$ 0.20*
	24	22.0 $\pm$ 1.80‡	13.6 $\pm$ 0.95†	8.30 $\pm$ 1.80	3.19 $\pm$ 1.25	1.25 $\pm$ 0.18	1.08 $\pm$ 0.07†

\*Source: Cambon et al. (5).

<sup>b</sup>AChE activity in controls measured on day 18 and day 19 of gestation, respectively.

\*Significantly different than control value at  $p < 0.05$ .

†Significantly different than control value at  $p < 0.001$ .

‡Significantly different than control value at  $p < 0.01$ .

(56) have been cholinergic in nature and have spontaneously subsided, generally within a 6-hr period. Clinical symptoms varying in severity with the amount of insecticide consumed and the age and general health of the exposed individual included dizziness, skeletal muscle weakness, epigastric cramping pain, diarrhea, excessive sweating, nausea, vomiting, nonreactive contracted pupils, blurred vision, dyspnea, and muscle fasciculation or convulsions. The causes of these manifestations are discussed in the section of this paper dealing with the mechanisms of toxicity.

## Accidental Poisonings

The death of a 20-year-old farmworker who had been loading granulated aldicarb (Temik 15G) into a hopper was reported by Lee and Ransdell (57). After initially working without protective equipment, the worker was provided with a vapor respirator, goggles, and paper coveralls and returned to his loading assignment without any observable signs or symptoms of problems. He was believed to have opened as many as 14 bags of Temik, and the empty packaging was burned about 200 feet from the loading site. Approximately 2 hr after beginning his first assignment as a pesticide loader, his body was found in an adjacent field. Initial postmortem procedures were performed without the knowledge that the victim had been working with Temik, and the immediate cause of death was determined to be massive crushing chest injuries from being run over by a tractor.

Upon belated arrival of information indicating possible pesticide exposure, liver, kidney, and skin samples were taken for residue analysis. Residues of aldicarb, aldicarb sulfoxide, and aldicarb sulfone were quantitated using high-performance liquid chromatography and an ultraviolet fluorescence technique. Detectable levels of total aldicarb derivatives (aldicarb, aldicarb sulfoxide, and aldicarb sulfone) were found in all tissues analyzed (Table 4). The notably high concentrations of the metabolites aldicarb sulfoxide and aldicarb sulfone observed in the renal tissue were reported to be evidence that excretion was already occurring at the time of death. Lee and Ransdell estimated the body burden,

based upon total water distribution, to be 18.2 mg aldicarb, or 0.275 mg/kg body weight, and concluded that the toxicological analyses and circumstances surrounding the accident indicated that aldicarb intoxication was a contributing factor. Since the victim was calculated to have nearly three times the level of blood and tissue aldicarb known to produce cholinergic symptoms in man, it was speculated that physical incapacitation due to aldicarb exposure may have rendered him unable to perform the necessary coordinated voluntary neuromuscular actions that might have prevented his death.

Two incidences of suspected carbamate poisoning in Nebraska in 1977 have been reported (54,55). In both instances, all individuals experiencing the symptoms of carbamate intoxication had just eaten hydroponically grown cucumbers purchased at the same market. In the first reported incident, nine residents of a Nebraska town suffered the onset of acute illness within 15 min to 2.25 hr following ingestion of hydroponic cucumbers produced in the same local greenhouse. The nine cases of illness, all experienced in a 2-week period, were characterized by the following symptoms: diarrhea, vomiting, excessive perspiration, blurred vision, abdominal pain, dyspnea, muscle fasciculation, nausea, and in some cases, headache and/or loss of function of arms and legs. Convulsions occurred in one case. The victims, three males and six females, ranged in age from 7 to 80 years and experienced symptoms of the sudden illness for 4 to 12 hr. All recovered quickly and completely. Hematological and chemical analyses revealed results with normal ranges, but cholinesterase levels were not determined. Analysis of the remaining portions of the uneaten cucumbers indicated that a carbamate was present, but the precise species could not be identified.

In 1978, five individuals became seriously ill after ingesting hydroponic cucumbers grown in the same greenhouse that had been involved in the 1977 poisonings (54,55). Onset of symptoms occurred within 30 to 60 min of ingestion of the cucumbers. The victims, three males and two females ranging in age from 6 to 49 years, manifested symptoms similar to those observed in the 1977 incident, and recovery in this instance was also rapid and complete. Analysis of cucumbers from the supermarkets where the suspect cucumbers had been purchased revealed the presence of aldicarb, as did sampling of materials from the greenhouse in which they were grown. Three of four cucumbers sampled contained aldicarb levels ranging from 6.6 to 10.7 ppm, and 1.8 ppm of aldicarb was identified in the water-nutrient solution that nourished the greenhouse cucumber plants.

Sexton (53) reported a case history of aldicarb poisoning experienced in a manufacturing plant. The victim of the poisoning was a foreman who ran a mechanical bagging machine for 1 day. Symptoms experienced included nausea, dizziness, depression, weakness, tightness of chest muscles, and decrease in plasma and RBC cholinesterase activity. The duration of symptoms was somewhat longer than 6 hr (exact duration unspecified),

Table 4. Residues of aldicarb and its metabolites in tissues of an exposed farmworker.<sup>a,b</sup>

Tissue	Aldicarb, ppm	Aldicarb sulfoxide, ppm	Aldicarb sulfone, ppm	Total aldicarb derivatives, ppm
Blood	0.028 <sup>c</sup>	0.108	0.374	0.482
Liver	0.013	0.058	0.116	0.187
Kidney	< 0.002	0.261	0.422	0.683
Skin (hand)	0.492	0.157	0.174	0.823
Skin (abdomen)	0.005	0.015	0.024 <sup>c</sup>	0.020
Skin (thigh)	0.168	0.126	0.083	0.377

<sup>a</sup> Source: Adapted from Lee and Ransdell (57).

<sup>b</sup> No explanation provided for differences in reported minimal detectable levels.

<sup>c</sup> Minimal detectable level.

and the subject returned to work the following day without symptoms.

In an experimental study with human subjects, three groups of four adult males, all in good health, were administered single oral doses of aldicarb (analytical grade, 99.2% pure) in water solutions corresponding to 0.1, 0.05, or 0.025 mg insecticide/kg body weight (56). Blood cholinesterase levels were monitored both before and after dosing, and symptoms resulting from the treatment were observed by physicians. Blood samples were collected from all subjects 18 hr and 1 hr before ingestion of the aldicarb, and at 1, 2, 4, and 6 hr after dosing. A maximum dose of 0.1 mg/kg body weight was selected based upon the 0.1 mg/kg body weight no-effect-level determined in the 2-year rat feeding study of Weil and Carpenter (33), while the other dosages selected for this experiment were one-half and one-fourth of the rat NOEL. Subjects receiving the 0.1 mg/kg dose manifested a variety of cholinergic symptoms including malaise, weakness in the arms and legs, pupils contracted and nonreactive to light, epigastric cramping pain, sweating of hands and forehead, air hunger, frequent yawning, salivation, slurred speech, nausea, and vomiting. The aldicarb-induced cholinesterase depression was reported to be rapidly reversible, and by 6 hr after administration, all symptoms had disappeared and the subjects were reported to feel normal again.

The author noted that the observed reduction in cholinesterase activity appeared to be out of proportion to the nature of the symptoms; two of the subjects that received the 0.1 mg/kg dose had cholinesterase activity values as low as 25% (31% if 1-hr predosing values are used) of their pre-exposure level. It was concluded that the radiometric method employed for cholinesterase level analysis gives an average value for red cell and plasma cholinesterase and may therefore yield a different interpretation than the usual analytical procedures that measure true acetylcholinesterase from the red cell. Furthermore, statistical analysis of these data suggests a dose-related effect when the maximum pre-dose control value is considered, leading to the assumption that at lower levels of ingestion (0.002–0.004 mg/kg), a significant depression of erythrocyte cholinesterase would probably not occur.

## Epidemiological Studies

Following the detection of aldicarb in well water samples in Suffolk County, New York, Varma et al. conducted a mail survey of families whose wells had been tested for aldicarb (58). The prospective respondents were requested to report every family member who had experienced any of a list of 20 general health problems, and women of reproductive age were to report the outcome of all pregnancies as well as the health of their children. A list of 25 randomly arranged neurological symptoms was also included for the respondents to identify family members who had experienced any of the symptoms.

The response rate to the more than 1500 question-

naires mailed was only about 20%. For a number of reasons, only households with aldicarb water concentrations  $\geq 8$  ppb were used for analysis of the results. Varma and his co-workers reported that the self-reported problems suggested an association between certain neurologic symptoms/syndromes and the concentration of aldicarb in the drinking water. The rate of spontaneous abortions (Table 5) was high among women consuming water with the highest aldicarb concentrations; however, it should be noted that the same group also had a high incidence of spontaneous abortions in the 7 years prior to the use of aldicarb in that farming region. A dose-related trend in the spontaneous abortion rates was observed for the 1975–1981 period; however, the abortion rates of 10 and 22.4% for the two study periods were not significantly different. The authors stated that selection and reporting bias could have influenced these findings, and that distortion at a survey response level as low as 20% could be a serious problem. The authors concluded that the findings of their study provided no conclusive evidence because: (1) the data obtained were based upon information provided by residents and not from medical records; (2) there was a lack of gynecological histories and data from all pregnant women; (3) the response rate to the questionnaires mailed was very low; (4) the contamination level used in the analysis was based in most instances on a single water sample, and therefore, might not be indicative of a long-term contamination level; and (5) the possible presence of other synthetic chemicals used on the soil could not be overruled.

## Mechanisms of Toxicity

The primary mode of aldicarb toxicity is cholinesterase inhibition. Carbamate insecticides are known to directly affect the enzyme acetylcholinesterase (AChE), which is associated with the outer surface of membranes. This results in a buildup of acetylcholine (ACh), which acts on the plasma membrane to produce the primary expression of neurotoxicity (59). It is commonly accepted that carbamates interfere with the ability of AChE to break down the chemical transmitter ACh at synaptic and myoneural junctions, although the precise biochemical mechanism for this interaction remains an object of discussion. It is known, however, that the same mechanism of action is evident in both target and nontarget organisms (6). Aldicarb and other carbamate insecticides further cause depression of other cholinesterases ("pseudocholinesterases") in the red blood cells and plasma of humans and other vertebrate species (4,17,21,28,29,36,37,56), but the degree of inhibition necessary to produce adverse effects in exposed subjects is speculative and the subject of current research.

Various cholinesterases have also been identified in the brain, liver, pancreas, intestine, heart, and skeletal muscle of mammals, and may be distinguished from one another, and from AChE ("true cholinesterases") by substrate and inhibitor specificity (6,52,59). Erythrocyte AChE is a more appropriate indicator of the level

Table 5. Reported pregnancies and spontaneous abortions among respondents to Suffolk County mail survey.<sup>a</sup>

	Aldicarb concentration ranges, ppb				Total/averages
	8-15	16-35	35-36	66+	
1968-1974 <sup>b</sup>					
Women responding	31	41	36	32	140
Women reporting pregnancies	4	8	7	6	25
Total pregnancies reported <sup>c</sup>	6	11	14	9	40
Spontaneous abortions <sup>d</sup>	1	0	1	2	4
Percent spontaneous abortions	16.7	0.0	7.1	22.2	10.0
1975-1981 <sup>e</sup>					
Women responding	32	46	33	29	140
Women reporting pregnancies	9	10	9	7	35
Total pregnancies reported <sup>c</sup>	17	19	18	13	67
Spontaneous abortions <sup>f</sup>	1	6 <sup>g</sup>	2	6 <sup>h</sup>	15
Percent spontaneous abortions	5.9	31.6	11.1	46.2	22.4

<sup>a</sup>Source: Modified from Varma et al. (58).

<sup>b</sup>Period prior to possible aldicarb exposure; concentration ranges merely denote geographic locations for comparison with 1975-1981 survey data.

<sup>c</sup>Includes multiple pregnancies in the same individual.

<sup>d</sup>Trend of spontaneous abortions, 1968-1974;  $Z = 0.0742$ ,  $p > 0.22$ .

<sup>e</sup>Period of possible aldicarb exposure.

<sup>f</sup>Trend of spontaneous abortions, 1975-1981;  $Z = 1.912$ ,  $p < 0.028$ .

<sup>g</sup>One woman had two spontaneous abortions (also one normal delivery in each of the 1968-1974 and 1975-1981 time periods).

<sup>h</sup>One woman had two spontaneous abortions in this period; one woman had one each in the 1968-1974 and 1975-1981 periods (both also had normal pregnancies).

of AChE in the central nervous system (CNS) than plasma AChE. Blood ChE generally becomes markedly depressed prior to the onset of cholinergic symptoms, and symptoms do not usually appear until the cholinesterase level reaches 25% of the pre-exposure value. A decrease of 60% in RBC AChE level warrants removal from the source of exposure (60).

Signs and symptoms of aldicarb intoxication are typically cholinergic and may be ameliorated by the administration of atropine sulfate (58,60). Since AChE is present in substantial excess at cholinergic synapses, 60 to 90% of the enzyme must be inhibited before the onset of cholinergic dysfunction (61). Symptoms of AChE inhibition and subsequent accumulation of ACh in nervous tissue and effector organs mimic the muscarinic, nicotinic, and CNS actions of ACh and may be categorized as follows (60):

1. *Muscarinic Signs.* The stimulation of muscarinic receptors (which are found primarily in the smooth muscle, the heart, and exocrine glands) results in the following symptoms:

- tightness in the chest and wheezing due to bronchoconstriction;
- increased bronchial secretions, salivation, lacrimation, and sweating;
- increased gastrointestinal tone, with consequent development of nausea, vomiting, abdominal cramps, diarrhea, and involuntary defecation;
- frequent contraction of smooth muscle of the bladder, resulting in involuntary urination;
- bradycardia that can progress to heart block;
- constriction of the pupils.

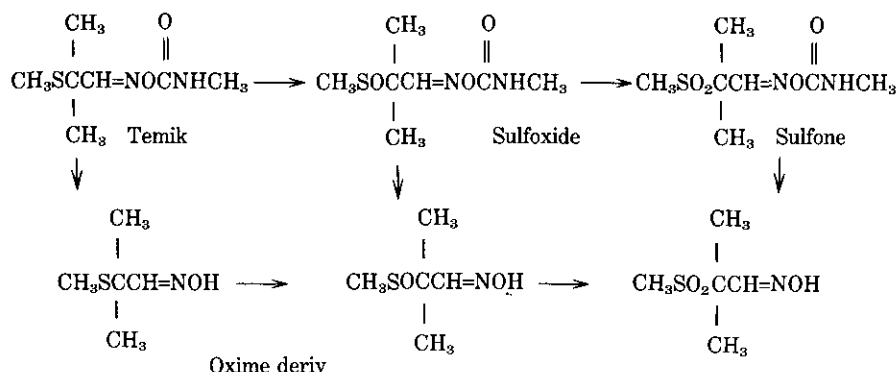
2.

*Nicotinic Signs.* The accumulation of ACh at the endings of motor nerves to skeletal muscle and autonomic ganglia results in the following symptoms:

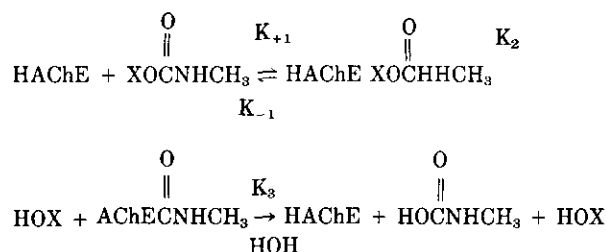
- Muscular effects, including easy fatigability and mild weakness, followed by involuntary twitching and cramps. Weakness affects the muscles involved in respiration and contributes to dyspnea, hypoxemia, and cyanosis.
- Nicotinic actions at autonomic ganglia may, in severe intoxication, mask some of the muscarinic effects. Thus, tachycardia caused by stimulation of sympathetic ganglia may override the usual bradycardia due to muscarinic action on the heart. Elevation of blood pressure and hyperglycemia also reflect nicotinic action at sympathetic ganglia.

Without going into a lengthy discussion of the morphology and function of the mammalian neuromuscular system, the transmission of electrical impulses between nerves and at myoneural junctions generally occurs through the release of chemical transmitters which bind with specific receptors on the postsynaptic terminal or motor end plate, respectively. As the chemical transmitter, acetylcholine in certain nerve synapses and at neuromuscular junctions binds to the receptor sites, an esterase (AChE) rapidly hydrolyzes the ACh into acetyl and choline fractions so that the stimulated nerves or muscles are not continually excited. Essentially, aldicarb and other cholinesterase inhibitors in some way prevent the breakdown of ACh and the subsequent return to a more normal or resting state for the nerve and/or muscle cells.

As opposed to the long-lasting cholinesterase inhibition associated with organophosphate pesticides, the carbamylation reactions involving the carbamates pro-

FIGURE 2. *In vivo* metabolism of aldicarb.

duce toxicity of a considerably shorter and more transient duration (61). Kuhr and Dorrough (6) suggest a formula for the reaction of *N*-methylcarbamate insecticides with acetylcholinesterase:



In this process, the enzyme HACHe reacts with the pesticide to form an intermediate complex which may: 1) dissociate to enzyme and carbamate, or 2) decompose into a stable carbamylated enzyme plus a leaving group, which is the parent phenol, naphthol or oxime (as in the case of aldicarb). The carbamylated enzyme is then hydrolyzed to generate free enzyme and methyl carbamic acid. It should be noted that this reaction sequence does not result in the destruction of AChE, but rather in the hydrolysis of the insecticide molecule itself.

During a normal nerve impulse, the AChE cleaves the ACh molecule. When a carbamate insecticide such as aldicarb enters the synapse, however, it competes with ACh for the active site on the enzyme. Since the decomposition to a carbamylated enzyme and leaving group, as well as the hydrolysis of the carbamylated enzyme, occur thousands of times slower when the active site is occupied with the carbamate than with ACh, the enzyme is essentially and effectively inhibited, and the normal cleavage of the ACh molecule by its esterase would be precluded. The greater the quantity of carbamate present, the greater the enzyme inhibition and concomitant reduction of ACh excitatory activity.

Reversal of this process may occur in two ways: 1) through dissociation of the pesticide-enzyme complex, and 2) a decarbamylation reaction, the half-life of which has been estimated to be 30 to 40 min (62,63). By these mechanisms, if insecticide exposure were not continual, recovery from anti-AChE activity would begin within

a few minutes of carbamylation and be complete (and the associated symptoms dissipated) within a matter of hours. This has been observed to be the case in human exposures (54-56).

The *in vivo* metabolism of aldicarb transforms the parent insecticide without activation into the sulfoxide and sulfone cholinesterase inhibiting forms (Fig. 2).

Further illumination of the mechanism of sensitivity to aldicarb was provided by Cambon et al. (52), who observed a significant inhibition of maternal and fetal cerebral AChE following exposure of the dams. These authors described the existence of three distinct isoenzymes that were involved, and reported that the treatment produced a significant lowering in the dams of the least mobile of the three (isoenzyme 1) and a decrease of isoenzymes 1 and 2 in the fetuses. Cambon and his co-workers suggested that the greater inhibition observed in fetal brain AChE activity in a previous study (5) might be due to the binding of the inhibitors for two of the isoenzymes in the fetuses as opposed to only the first isoenzyme in the mothers at the same dosage level, thereby indicating a greater affinity of the carbamate derivatives, such as aldicarb, for each respective fetal and maternal isoenzyme.

## Conclusions

The primary mode of toxic action of aldicarb is cholinesterase inhibition. Reports of one death and multiple accidental poisonings in humans indicate relatively rapid remission of symptoms, unless the exposure is so severe as to result in a life threatening situation and death. To date, aldicarb has been found to exhibit no carcinogenic activity, but the potential for *in vivo* transformation to the nitroso derivative (which is carcinogenic) has been suggested as a likely possibility. Recent positive mutagenicity studies suggest that this insecticide has mutagenic properties, but other negative studies make an unequivocal determination of mutagenic potential inappropriate at this time. No teratogenic or long-term toxic effects as a result of aldicarb exposure have been reported.

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